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Prevention, Pesticides
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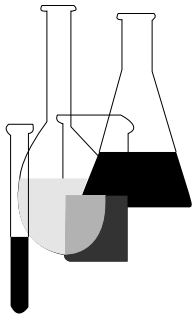
Health Effects Test Guidelines OPPTS 870.1000 Acute Toxicity Testing—Background



INTRODUCTION

This guideline is one of a series of test guidelines that have been developed by the Office of Prevention, Pesticides and Toxic Substances, United States Environmental Protection Agency for use in the testing of pesticides and toxic substances, and the development of test data that must be submitted to the Agency for review under Federal regulations.

The Office of Prevention, Pesticides and Toxic Substances (OPPTS) has developed this guideline through a process of harmonization that blended the testing guidance and requirements that existed in the Office of Pollution Prevention and Toxics (OPPT) and appeared in Title 40, Chapter I, Subchapter R of the Code of Federal Regulations (CFR), the Office of Pesticide Programs (OPP) which appeared in publications of the National Technical Information Service (NTIS) and the guidelines published by the Organization for Economic Cooperation and Development (OECD).

The purpose of harmonizing these guidelines into a single set of OPPTS guidelines is to minimize variations among the testing procedures that must be performed to meet the data requirements of the U. S. Environmental Protection Agency under the Toxic Substances Control Act (15 U.S.C. 2601) and the Federal Insecticide, Fungicide and Rodenticide Act (7 U.S.C. 136, *et seq.*).

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OPPTS 870.1000 Acute toxicity testing—background.

(a) **Scope**—(1) **Applicability.** This guideline is intended to meet testing requirements of both the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) (7 U.S.C. 136, *et seq.*) and the Toxic Substances Control Act (TSCA) (15 U.S.C. 2601).

(2) [Reserved]

(b) **Purpose.** The Agency considers the evaluation of toxicity following short term exposure to a chemical to be an integral step in the assessment of its toxic potential under the regulatory framework of its pesticide and toxic substances programs. In the assessment and evaluation of the toxic characteristics of a substance, acute toxicity is generally performed by the probable route of exposure in order to provide information on health hazards likely to arise from short-term exposure by that route. For pesticides, the short-term toxicity testing battery consists of acute toxicity tests by the oral, dermal, and inhalation routes; skin and eye irritation testing; and testing for dermal sensitization. Data from an acute study may serve as a basis for hazard categorization, labeling, or child-resistant packaging and may also serve to designate pesticides which may be applied only by certified applicators. It is also an initial step in establishing a dosage regimen in subchronic and other studies and may provide information on absorption and the mode of toxic action of a substance. An evaluation of acute toxicity data should include the relationship, if any, between the exposure of animals to the test substance and the incidence and severity of all abnormalities, including behavioral and clinical abnormalities, the reversibility of observed abnormalities, gross lesions, body weight changes, effects on mortality, and any other toxic effects.

(c) **History**—(1) **Acute toxicity test guidelines.** Test guidelines for acute toxicity were first published by the Agency in October 1982 as part of Subdivision F of the Pesticide Assessment Guidelines for the Office of Pesticide Programs (OPP) (see paragraph (f)(4) of this guideline) and in 40 CFR part 797 in September 1985 for the Office of Toxic Substances (OPPTS).

(2) **Rejection rate analysis.** In 1993, as part of its Pesticide Rejection Rate Analysis, Agency and industry scientists met to perform a guideline-by-guideline review of toxicology studies including acute toxicity studies. The purpose of this guideline-by-guideline review was to identify those factors that most frequently cause toxicology studies required for pesticide reregistration to be rejected. The results were published as the *Pesticide Reregistration Rejection Rate Analysis: Toxicology* (see paragraph (f)(5) of this guideline). In 1995, representatives from the Agency met with the American Crop Protection Association (ACPA), the Chemical Producers and Distributors Association (CPDA), the Chemical Manufacturers Association (CMA), Health Canada, and the California Department of Pesticide Regulation (CDPR) to discuss acceptable methods for the conduct of acute

toxicity studies. The discussions of this meeting were incorporated into a preliminary Registration Division document titled *Conduct of Acute Toxicity Studies* (see paragraph (f)(6) of this guideline). These documents supplement the acute toxicology guidelines in Subdivision F.

(3) **Guideline harmonization.** The Series 870 Health Effects test guidelines have been harmonized between OPP and OPPTS and, where possible, with OECD test guidelines. Scientific considerations from both of the analyses described in paragraph (c)(2) of this guideline have been incorporated into the revised test guidelines.

(d) **Approaches to the determination of acute toxicity.** (1) At present, the evaluation of chemicals for acute toxicity is necessary for the protection of public health and the environment. The Agency supports measures dedicated to reducing the use of animals in toxicity testing. When animal testing is required for this purpose, testing should be done in ways that minimize numbers of animals used and that take full account of their welfare. To this end, when conducting a test, the Agency stresses the simultaneous monitoring of several endpoints of toxicity in animals in a single acute study including sublethal effects as well as lethality. Dosed animals are observed for abnormal behavioral manifestations such as increased salivation or muscular incoordination, in addition to the recovery from these effects during the observation period. Both dead and surviving animals are necropsied to evaluate gross anatomical evidence of organ toxicity. In selected cases, additional testing may be justified to better characterize the kinds of abnormalities that have been found in the organs of the necropsied animals. These sound, scientific practices represent some of the means which maximize the utility of the data obtained from a limited number of test animals to achieve a balance between protecting humans and the environment, and the welfare and utilization of laboratory animals.

(2) EPA recommends the following means to reduce the number of animals used to evaluate acute effects of chemical exposure while preserving its ability to make reasonable judgements about safety:

(i) Use of data from structurally related substances or mixtures. In order to minimize the need for animal testing for acute effects, the Agency encourages the review of existing acute-toxicity information on chemical substances that are structurally related to the agent under investigation. In certain cases, it may be possible to obtain enough information to make preliminary hazard evaluations that may reduce the need for further animal testing for acute effects. Similarly, mixtures or formulated products that are substantially similar to well-characterized mixtures or products may not need additional testing if there are sufficient bridging data available for meaningful extrapolation. In those cases, classification would be extrapolated from the mixture already tested.

(ii) Use of appropriate alternative test protocols when available. Thus, for example, acute oral toxicity testing may be performed using the Fixed Dose Method (OECD Guideline 420, see paragraph (f)(1) of this guideline), or the Acute Toxic Class Method (OECD Guideline 423, see paragraph (f)(2) of this guideline), or the Up-and-Down Method (OECD Guideline 425, see paragraph (f)(3) of this guideline). Abbreviated methods are not yet available through OECD for acute toxicity by other routes of exposure.

(iii) Weight of evidence approaches to dermal and ocular irritation. Several factors should be considered in determining the corrosion and irritation potential of chemicals before testing is undertaken. Existing human experience and data and animal observations and data should be the first line of analysis, as it gives information directly referable to effects on the skin. In some cases, enough information may be available from structurally related compounds to make classification decisions. Likewise, pH extremes (pH <2 or >11.5) may indicate dermal effects, especially when buffering capacity is known, although the correlation is not perfect. Generally, such agents are expected to produce significant effects on the skin. It also stands to reason that if a chemical is extremely toxic by the dermal route, a dermal irritation/corrosion study may not be needed. Likewise, if there is a lack of any dermal reaction at the limit dose (2,000 mg/kg) in an acute toxicity study (for which observations of dermal reactions were made), a dermal irritation/corrosion study again may not be needed. It should be noted, however, that often acute dermal toxicity and dermal irritation/corrosion testing are performed in different species that may differ in sensitivity. *In vitro* alternatives that have been validated and accepted may also be used to help make classification decisions.

(iv) All of the available information on a chemical should be used in determining the need for *in vivo* dermal irritation testing. Although information might be gained from the evaluation of single parameters within a tier (e.g., caustic alkalies and acids with extreme pH (pH <2 or >11.5) should be considered as dermal corrosives), there is merit in considering the totality of existing information and making an overall weight of evidence determination. This is especially true when there is information available on some but not all parameters.

(v) Use of limit testing. For chemicals judged to be relatively nontoxic, a single group of animals is given a large dose of the agent. If no lethality is demonstrated, no further testing is pursued. The substance is classified in hazard categories according to the limit dose used. (See the following paragraph for a discussion of toxicity categories under FIFRA).

(e) **Regulatory applications under FIFRA.** (1) Precautionary labeling provides the pesticide user with a general idea of the potential toxicity, irritation and sensitization hazard associated with the use of a pesticide

(see EPA Label Review Manual (paragraph (f)(7) of this guideline) and 40 CFR Part 156—Labeling Requirements for Pesticides and Devices). Precautionary labeling also identifies the precautions necessary to avoid exposure as well as any personal protective equipment which should be used when handling a pesticide and statements of practical treatment in case of accidental exposure. The United States is an active participant in negotiations to develop a globally harmonized system for classification and labeling. Planning for the globally harmonized system will be completed in the year 2000 with implementation to be phased in after planning is completed. This section describes the current system in place for pesticides in the United States and will be revised and updated when the globally harmonized system is fully implemented.

(2) Precautionary labeling which includes the signal word, personal protective equipment, hazard symbol, and statements of practical treatment is normally determined by six acute toxicity studies and product composition. The acute oral, acute dermal and acute inhalation studies are used to determine the LD_{50} of a product via the designated route of exposure. The primary eye irritation and primary skin irritation studies measure the severity of irritation or corrosivity caused by a product. The dermal sensitization study determines whether a product is capable of causing an allergic reaction. With the exception of the dermal sensitization study, each acute toxicity study is assigned a toxicity category as defined in the table below. All products falling into toxicity categories I–IV must bear a signal word and in some cases warning symbols.

(3) Personal Protective Equipment. Personal protective equipment which includes use of protective clothing, chemical resistant gloves, protective eye gear, and respiratory protective devices, is determined by the results of six acute toxicity studies according to toxicity category (see table). The degree of protection required is graded according to the degree of acute toxicity and the hazard classification category of the chemical or product. These requirements are set forth in 40 CFR 170.240 in the Worker Protection Standard.

(4) Restricted entry intervals. Agricultural products must display a restricted entry interval. A restricted entry interval is the time immediately following a pesticide application during which entry into the treated area is restricted. Restricted entry intervals are based on the most severe acute toxicity category assigned to the acute dermal, eye irritation and skin irritation data for all of the active ingredients in a pesticide product. The duration of restricted entry intervals is based on the severity of toxicity, with products classified in category I requiring intervals of 48 hours or more and products classified in category III or IV requiring intervals of 12 hours.

(5) Child-resistant packaging. FIFRA establishes standards with respect to pesticide packaging of products intended for use in residential

settings in order to protect children or adults from serious illness or injury resulting from accidental ingestion or contact with pesticides. Criteria for which pesticides must be distributed or sold in child-resistant packaging are based on classification according to the toxicity categories set forth in the table.

(6) Restricted use pesticide. The Agency determines whether a pesticide must be applied under the direct supervision of a certified applicator. Such clarification for restricted use is based upon consideration of toxicity data, including acute toxicity, exposure, and intended use.

(7) Biochemical pest control agents are tested in a special tiered progression. The technical grade biochemical pest control agent is always characterized by acute toxicity tests. However, because of their nontoxic mode of action against the target pest, further testing of the biochemical pest control agent is normally not required. Microbial pest control agents are tested using the OPPTS Harmonized Test Guidelines Series 885, Microbial Pesticide Test Guidelines, for pathogenicity/infectivity. In addition, all formulations of microbial pest control agents are tested for precautionary labeling using acute toxicity tests in the OPPTS Harmonized Test Guidelines Series 870, Health Effects Test Guidelines.

Toxicity Categories

Study	CategoryI	Category II	Category III	Category IV
Acute Oral	Up to and including 50 mg/kg	>50 through 500 mg/kg	>500 through 5000 mg/kg	>5000 mg/kg
Acute Dermal	Up to and including 200 mg/kg	>200 through 2000 mg/kg	>2000 through 5000 mg/kg	>5000 mg/kg
Acute Inhalation	Up to and including 0.05 mg/liter	>0.05 through 0.5 mg/liter	>0.5 through 2 mg/liter	>2 mg/liter
Eye Irritation	Corrosive (irreversible destruction of ocular tissue) or corneal involvement or irritation persisting for more than 21 days	Corneal involvement or irritation clearing in 8-21 days	Corneal involvement or irritation clearing in 7 days or less	Minimal effects clearing in less than 24 hours
Skin irritation	Corrosive (tissue destruction into the dermis and/or scarring)	Severe irritation at 72 hours (severe erythema or edema)	Moderate irritation at 72 hours (moderate erythema)	Mild or slight irritation (no irritation or slight erythema)

Study	Study results	Study results
Dermal Sensitization	Product is a sensitizer or is positive for sensitization	Product is not a sensitizer or is negative for sensitization

(f) **References.** The following references should be consulted for additional background information on this test guideline.

(1) Organization for Economic Cooperation and Development, OECD Guidelines for Testing of Chemicals. Guideline 420: Acute Oral Toxicity-Fixed Done Method. Adopted: July 17, 1992.

(2) Organization for Economic Cooperation and Development, OECD Guidelines for Testing of Chemicals. Guideline 423: Acute Oral Toxicity-Acute Toxic Class Method. Adopted: March 22, 1996.

(3) Organization for Economic Cooperation and Development, OECD Guidelines for Testing of Chemicals. Guideline 425: Acute Oral Toxicity–Up-and-Down Method. Approved: June 1998.

(4) U.S. Environmental Protection Agency. Pesticide Assessment Guidelines, Subdivision F: Health Effects. EPA report 540/09–82–025, October 1982.

(5) U.S. Environmental Protection Agency. Pesticide Reregistration Rejection Rate Analysis: Toxicology. EPA report 738–R–93–004. July 1993.

(6) U.S. Environmental Protection Agency. *Conduct of Acute Toxicity Studies.* EPA report 737–R–97–002. September 1997.

(7) U.S. Environmental Protection Agency. *Label Review Manual* 2nd Edition. EPA report 737–B–96–001. December 1996.